



RAW SEQUENCE LISTING
PATENT APPLICATION: US/09/647,522

DATE: 03/18/2002
TIME: 15:20:39

Input Set : A:\Nagai h.app
Output Set: N:\CRF3\03182002\I647522.raw

Law Secretary
7/31/03
P.

OK

3 <110> APPLICANT: NAGAI, Hiroshi
4 NAKAJIMA, Terumi
6 <120> TITLE OF INVENTION: NOVEL HEMOLYTIC ACTIVE PROTEINS AND GENES ENCODING THE
7 SAME
9 <130> FILE REFERENCE: PCTJP9901607NAGAIHiroshietal 183049264
11 <140> CURRENT APPLICATION NUMBER: US/09/647,522
12 <141> CURRENT FILING DATE: 2000-10-02
14 <150> PRIOR APPLICATION NUMBER: PCT/JP99/01607
15 <151> PRIOR FILING DATE: 1999-03-30
17 <150> PRIOR APPLICATION NUMBER: JP 10/88569
18 <151> PRIOR FILING DATE: 1998-04-01
20 <160> NUMBER OF SEQ ID NOS: 18
22 <170> SOFTWARE: PatentIn Ver. 2.0
24 <210> SEQ ID NO: 1
25 <211> LENGTH: 14
26 <212> TYPE: PRT
27 <213> ORGANISM: Carybdea rastonii
29 <220> FEATURE:
30 <223> OTHER INFORMATION: This amino acid residue sequence corresponds to
31 amino acid residue positions 56-69 of SEQ ID NO:5.
33 <400> SEQUENCE: 1
34 Gly Glu Ile Gln Thr Lys Pro Asp Arg Val Gly Gln Ala Thr
35 1 5 10
38 <210> SEQ ID NO: 2
39 <211> LENGTH: 18
40 <212> TYPE: PRT
41 <213> ORGANISM: Carybdea rastonii
43 <220> FEATURE:
44 <223> OTHER INFORMATION: This amino acid residue sequence corresponds to
45 amino acid residue positions 250-267 of SEQ ID
46 NO:5.
48 <400> SEQUENCE: 2
49 Gly Asn Ala Glu His Val Ala Ser Ala Val Glu Asn Ala Asn Arg Val
50 1 5 10 15
52 Asn Lys
56 <210> SEQ ID NO: 3
57 <211> LENGTH: 15
58 <212> TYPE: PRT
59 <213> ORGANISM: Carybdea rastonii
61 <220> FEATURE:
62 <223> OTHER INFORMATION: This amino acid residue sequence corresponds to
63 amino acid residue positions 363-377 of SEQ ID
64 NO:5.

ENTERED

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66 <400> SEQUENCE: 3

67 Met Ser Asp Gly Phe Tyr Thr Met Glu Asn Ser Asp Arg Arg Lys

68 1 5 10 15

71 <210> SEQ ID NO: 4

72 <211> LENGTH: 1610

73 <212> TYPE: DNA

74 <213> ORGANISM: Carybdea rastonii

76 <220> FEATURE:

77 <221> NAME/KEY: CDS

78 <222> LOCATION: (28)..(1380)

80 <220> FEATURE:

81 <221> NAME/KEY: protein_bind

82 <222> LOCATION: (1381)..(1610)

84 <400> SEQUENCE: 4

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86 Met Ile Leu Lys His Leu Pro Trp Leu

87 1 5

89 ttt att gtc ctt gca att act tct gca aaa cat ggc aaa cgc tct gat 102

90 Phe Ile Val Leu Ala Ile Thr Ser Ala Lys His Gly Lys Arg Ser Asp

91 10 15 20 25

93 gtc aat tct tta ctt act aag gta gaa act gcc tta aaa gaa gct tct 150

94 Val Asn Ser Leu Leu Thr Lys Val Glu Thr Ala Leu Lys Glu Ala Ser

95 30 35 40

97 ggt agc aac gag gct gct ctt gag gct tta gag ggc tta aaa gga gag 198

98 Gly Ser Asn Glu Ala Ala Leu Glu Ala Leu Glu Gly Leu Lys Gly Glu

99 45 50 55

101 atc cag aca aaa cca gac cga gtt gga caa gcc aca aaa atc ctt gga 246

102 Ile Gln Thr Lys Pro Asp Arg Val Gly Gln Ala Thr Lys Ile Leu Gly

103 60 65 70

105 tct gtc gga tca gct cta gga aaa tta aat tct gga gat gca acc aaa 294

106 Ser Val Gly Ser Ala Leu Gly Lys Leu Asn Ser Gly Asp Ala Thr Lys

107 75 80 85

109 atc att tct ggt tgc ctc gac att gtt gca gga att gca aca act ttt 342

110 Ile Ile Ser Gly Cys Leu Asp Ile Val Ala Gly Ile Ala Thr Thr Phe

111 90 95 100 105

113 gga ggc cct gtc ggg atg gga atc gga gcc gta gct tct ttt gtt tct 390

114 Gly Gly Pro Val Gly Met Gly Ile Gly Ala Val Ala Ser Phe Val Ser

115 110 115 120

117 tca att cta tca ttg ttt act gga agc tca gca aag aac tca gtt gct 438

118 Ser Ile Leu Ser Leu Phe Thr Gly Ser Ser Ala Lys Asn Ser Val Ala

119 125 130 135

121 gcc gtt att gat aga gct tta agc aag cat cgc gat gag gcc atc caa 486

122 Ala Val Ile Asp Arg Ala Leu Ser Lys His Arg Asp Glu Ala Ile Gln

123 140 145 150

125 aga cat gca gca ggt gcc aag aga gat ttt gct gaa tca tct gca ttc 534

126 Arg His Ala Ala Gly Ala Lys Arg Asp Phe Ala Glu Ser Ser Ala Phe

127 155 160 165

129 att cag gtc atg aaa cag cag tcc aat ctt aca gat agc gac cta agt 582

130 Ile Gln Val Met Lys Gln Gln Ser Asn Leu Thr Asp Ser Asp Leu Ser

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131	170				175				180				185				
133	atc	att	gca	gcg	aat	gtt	cct	gtt	tat	aaa	ttt	agt	aat	ttt	atc	gga	630
134	Ile	Ile	Ala	Ala	Asn	Val	Pro	Val	Tyr	Lys	Phe	Ser	Asn	Phe	Ile	Gly	
135					190					195				200			
137	cag	ttg	gag	agc	aga	att	tcc	caa	ggc	gca	gca	act	acc	agt	ctt	agc	678
138	Gln	Leu	Glu	Ser	Arg	Ile	Ser	Gln	Gly	Ala	Ala	Thr	Thr	Ser	Leu	Ser	
139				205					210					215			
141	gat	gca	aag	aga	gcc	gtt	gac	ttc	att	ctg	ctc	tat	tgt	caa	ctt	gta	726
142	Asp	Ala	Lys	Arg	Ala	Val	Asp	Phe	Ile	Leu	Leu	Tyr	Cys	Gln	Leu	Val	
143			220					225						230			
145	gtc	atg	aga	gaa	acc	ttg	ctg	gtc	gac	ttg	gct	att	ctc	tac	agg	aaa	774
146	Val	Met	Arg	Glu	Thr	Leu	Leu	Val	Asp	Leu	Ala	Ile	Leu	Tyr	Arg	Lys	
147		235					240						245				
149	gga	aat	gca	gaa	cac	gtg	gca	agt	gct	gtg	gaa	aac	gct	aat	agg	gta	822
150	Gly	Asn	Ala	Glu	His	Val	Ala	Ser	Ala	Val	Glu	Asn	Ala	Asn	Arg	Val	
151	250				255					260				265			
153	aac	aaa	gag	cta	gct	gct	gat	acc	cta	gat	ttt	ctt	cat	aaa	ttg	att	870
154	Asn	Lys	Glu	Leu	Ala	Ala	Asp	Thr	Leu	Asp	Phe	Leu	His	Lys	Leu	Ile	
155				270					275					280			
157	cct	gaa	caa	gca	ttg	ata	ggt	gca	gtt	tat	cat	cca	att	tct	gcc	tct	918
158	Pro	Glu	Gln	Ala	Leu	Ile	Gly	Ala	Val	Tyr	His	Pro	Ile	Ser	Ala	Ser	
159			285					290						295			
161	gaa	act	agc	aaa	gca	ata	tta	aat	tac	acg	aaa	tac	ttt	gga	gtt	cca	966
162	Glu	Thr	Ser	Lys	Ala	Ile	Leu	Asn	Tyr	Thr	Lys	Tyr	Phe	Gly	Val	Pro	
163			300					305						310			
165	gat	gtt	ccc	cgt	cct	att	gga	aac	cgc	aga	tac	aaa	ttt	aca	aat	agt	1014
166	Asp	Val	Pro	Arg	Pro	Ile	Gly	Asn	Arg	Arg	Tyr	Lys	Phe	Thr	Asn	Ser	
167		315					320					325					
169	tac	tgg	aat	acc	tac	agt	ata	tgc	agt	gag	gct	tac	atg	gga	aat	tac	1062
170	Tyr	Trp	Asn	Thr	Tyr	Ser	Ile	Cys	Ser	Glu	Ala	Tyr	Met	Gly	Asn	Tyr	
171	330				335					340				345			
173	atg	ttc	aga	ggc	tgt	tct	aac	gtt	cgg	aat	cca	aat	atc	agg	gta	tcc	1110
174	Met	Phe	Arg	Gly	Cys	Ser	Asn	Val	Arg	Asn	Pro	Asn	Ile	Arg	Val	Ser	
175				350					355					360			
177	aaa	atg	tct	gat	ggg	ttt	tac	acc	atg	gag	aat	agc	gat	cgg	agg	aag	1158
178	Lys	Met	Ser	Asp	Gly	Phe	Tyr	Thr	Met	Glu	Asn	Ser	Asp	Arg	Arg	Lys	
179			365					370						375			
181	ttg	tat	atc	acc	aag	cat	gac	caa	gga	tgg	gga	tgg	ggt	act	ttg	gat	1206
182	Leu	Tyr	Ile	Thr	Lys	His	Asp	Gln	Gly	Trp	Gly	Trp	Gly	Thr	Leu	Asp	
183			380					385						390			
185	gag	gat	cca	ggt	gac	caa	ggc	cat	atg	agg	ttc	att	cct	ttg	aga	cat	1254
186	Glu	Asp	Pro	Gly	Asp	Gln	Gly	His	Met	Arg	Phe	Ile	Pro	Leu	Arg	His	
187		395				400						405					
189	ggg	aag	tat	atg	gta	agc	tct	aag	agg	tgg	ccc	aac	tgg	ttc	atg	tat	1302
190	Gly	Lys	Tyr	Met	Val	Ser	Ser	Lys	Arg	Trp	Pro	Asn	Trp	Phe	Met	Tyr	
191	410				415					420				425			
193	atg	gaa	tca	agt	gcc	agt	ggc	tac	att	cgc	agc	tgg	gaa	aat	aat	cca	1350
194	Met	Glu	Ser	Ser	Ala	Ser	Gly	Tyr	Ile	Arg	Ser	Trp	Glu	Asn	Asn	Pro	
195				430					435					440			

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197 gga cct caa gga cat tgg agt ata aca taa ttaaagagga atcaacaatg      1400
198 Gly Pro Gln Gly His Trp Ser Ile Thr
199          445                      450
201 tcccaaaggc atacgaatat aagacatcaa acgaatgcag tacttaaagt gcacacttgt 1460
203 atttctacat aggatgtcgt catgaaagtc cataaaccat ccagcggact aatttcatat 1520
205 taaacattaa tgtttcctta taatgcattt tcatgaaatc tctattgtga catttcaaga 1580
207 ggatatgttt gaaagaaaca aaaaaaaaaa                      1610
210 <210> SEQ ID NO: 5
211 <211> LENGTH: 450
212 <212> TYPE: PRT
213 <213> ORGANISM: Carybdea rastonii
215 <400> SEQUENCE: 5
216 Met Ile Leu Lys His Leu Pro Trp Leu Phe Ile Val Leu Ala Ile Thr
217   1           5           10           15
219 Ser Ala Lys His Gly Lys Arg Ser Asp Val Asn Ser Leu Leu Thr Lys
220           20           25           30
222 Val Glu Thr Ala Leu Lys Glu Ala Ser Gly Ser Asn Glu Ala Ala Leu
223           35           40           45
225 Glu Ala Leu Glu Gly Leu Lys Gly Glu Ile Gln Thr Lys Pro Asp Arg
226           50           55           60
228 Val Gly Gln Ala Thr Lys Ile Leu Gly Ser Val Gly Ser Ala Leu Gly
229   65           70           75           80
231 Lys Leu Asn Ser Gly Asp Ala Thr Lys Ile Ile Ser Gly Cys Leu Asp
232           85           90           95
234 Ile Val Ala Gly Ile Ala Thr Thr Phe Gly Gly Pro Val Gly Met Gly
235           100          105          110
237 Ile Gly Ala Val Ala Ser Phe Val Ser Ser Ile Leu Ser Leu Phe Thr
238           115          120          125
240 Gly Ser Ser Ala Lys Asn Ser Val Ala Ala Val Ile Asp Arg Ala Leu
241           130          135          140
243 Ser Lys His Arg Asp Glu Ala Ile Gln Arg His Ala Ala Gly Ala Lys
244 145          150          155          160
246 Arg Asp Phe Ala Glu Ser Ser Ala Phe Ile Gln Val Met Lys Gln Gln
247           165          170          175
249 Ser Asn Leu Thr Asp Ser Asp Leu Ser Ile Ile Ala Ala Asn Val Pro
250           180          185          190
252 Val Tyr Lys Phe Ser Asn Phe Ile Gly Gln Leu Glu Ser Arg Ile Ser
253           195          200          205
255 Gln Gly Ala Ala Thr Thr Ser Leu Ser Asp Ala Lys Arg Ala Val Asp
256           210          215          220
258 Phe Ile Leu Leu Tyr Cys Gln Leu Val Val Met Arg Glu Thr Leu Leu
259 225          230          235          240
261 Val Asp Leu Ala Ile Leu Tyr Arg Lys Gly Asn Ala Glu His Val Ala
262           245          250          255
264 Ser Ala Val Glu Asn Ala Asn Arg Val Asn Lys Glu Leu Ala Ala Asp
265           260          265          270
267 Thr Leu Asp Phe Leu His Lys Leu Ile Pro Glu Gln Ala Leu Ile Gly
268           275          280          285
270 Ala Val Tyr His Pro Ile Ser Ala Ser Glu Thr Ser Lys Ala Ile Leu

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```

271      290      295      300
273 Asn Tyr Thr Lys Tyr Phe Gly Val Pro Asp Val Pro Arg Pro Ile Gly
274 305      310      315      320
276 Asn Arg Arg Tyr Lys Phe Thr Asn Ser Tyr Trp Asn Thr Tyr Ser Ile
277      325      330      335
279 Cys Ser Glu Ala Tyr Met Gly Asn Tyr Met Phe Arg Gly Cys Ser Asn
280      340      345      350
282 Val Arg Asn Pro Asn Ile Arg Val Ser Lys Met Ser Asp Gly Phe Tyr
283      355      360      365
285 Thr Met Glu Asn Ser Asp Arg Arg Lys Leu Tyr Ile Thr Lys His Asp
286      370      375      380
288 Gln Gly Trp Gly Trp Gly Thr Leu Asp Glu Asp Pro Gly Asp Gln Gly
289 385      390      395      400
291 His Met Arg Phe Ile Pro Leu Arg His Gly Lys Tyr Met Val Ser Ser
292      405      410      415
294 Lys Arg Trp Pro Asn Trp Phe Met Tyr Met Glu Ser Ser Ala Ser Gly
295      420      425      430
297 Tyr Ile Arg Ser Trp Glu Asn Asn Pro Gly Pro Gln Gly His Trp Ser
298      435      440      445
300 Ile Thr
301      450

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304 <210> SEQ ID NO: 6

305 <211> LENGTH: 19

306 <212> TYPE: DNA

307 <213> ORGANISM: Artificial Sequence

309 <220> FEATURE:

310 <223> OTHER INFORMATION: Description of Artificial Sequence: Degenerate PCR

311 primer, 7-F, used in the cloning of the partial

312 cDNA of the hemolytic active protein of Carybdea

313 rastonii

315 <220> FEATURE:

316 <221> NAME/KEY: unsure

317 <222> LOCATION: (12)

318 <223> OTHER INFORMATION: n = inosine

320 <220> FEATURE:

321 <221> NAME/KEY: unsure

322 <222> LOCATION: (18)

323 <223> OTHER INFORMATION: n = inosine

325 <400> SEQUENCE: 6

OK
W-F-326 garathcara cnaarccng

19

329 <210> SEQ ID NO: 7

330 <211> LENGTH: 19

331 <212> TYPE: DNA

332 <213> ORGANISM: Artificial Sequence

334 <220> FEATURE:

335 <221> NAME/KEY: unsure

336 <222> LOCATION: (2)

337 <223> OTHER INFORMATION: n = inosine

339 <220> FEATURE:

Use of n and/or Xaa has been detected in the Sequence Listing.
Review the Sequence Listing to insure a corresponding
explanation is presented in the <220> to <223> fields of
each sequence using n or Xaa.

VERIFICATION SUMMARY

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L:11 M:270 C: Current Application Number differs, Replaced Application Number
L:12 M:271 C: Current Filing Date differs, Replaced Current Filing Date
L:326 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:6
L:351 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:7
L:381 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:8
L:411 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:9
L:436 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:10
L:461 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:11
L:486 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:12
L:511 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:13